Ghrelin mediates stress-induced food-reward behavior in mice

Chuang et al 2011 | L3: Love, Lust, Labor
Agenda

- Introduction
  - What is Ghrelin?
  - Previous Models
  - New model
- Methods
- Results
- Discussion
- Conclusion
- Implications
Stress Eating: Do-Nut Eat that!

- People experience altered feeding behavior upon stress
  - Increases in highly palatable food AKA “comfort food”
- Also found in people with major depressive disorder with “atypical” subtype
- Complex eating patterns stimulated by stress and depression leads to weight gain
- What’s the reason for this??
Limitation of Models

- Animal models developed to explore stress-induced and depression-associated eating behaviors
  - However, no insight into the motivations and decision-making processes at work
- Limited identification of the actual molecular substrates involved in stress eating
- Current research suggests association between stress and signaling by ghrelin, a peptide hormone produced in stomach & gastrointestinal tract
What is Ghrelin?

- 28-amino acid peptide produced in stomach’s X/A like cells
- Acts primarily through its growth hormone secretagogue receptor, GHS-R
- Hormone that stimulates appetite and adiposity
  - (Before meals) = ↑↑↑ Ghrelin
  - (During/after meals) = ↓↓↓ Ghrelin
- Roles
  - MAIN → Energy Balance
  - Promoting food intake
  - Regulation of gastric and pancreatic activity
  - Stimulation of growth hormone release and cardiovascular activities
Mechanism Under Ghrelin’s Orexigenic Effect

1) Ghrelin binds to growth hormone secretagogue receptor (GHS-R 1a)

2) Increased expression of:
   a) Forkhead box O1 (FOXO1)
   b) Phosphorylated cAMP response-element binding protein (pCREB)
   c) Hypothalamic homeobox domain transcription factor (BSX)

3) Stimulation of expression in arcuate nucleus of hypothalamus
   a) Agouti-related peptide (Agrp)
   b) Neuropeptide Y (Npy)

Ghrelin → hypothalamus “appetite regulator” → increase in appetite

Result: Ghrelin upregulates expression of Agrp and Npy in the ARC of the hypothalamus to trigger hunger
Models: How Ghrelin promotes food intake

- Peripheral ghrelin injection and prolonged caloric restriction to induce:
  - Conditioned Place Preference Task (CCP) for high fat diets (HFD)
  - Operant nose poking for HFD
  - Operant lever pressing for sucrose

- Growth Hormone Secretagogue Receptor (GHSRs)
  - Antagonist administration and genetic deletion of GHSRs prevent caloric restriction-associated CPP for HFD and caloric restriction-associated operant pressing for sucrose
How Ghrelin promotes food intake

- **Rats** → Ghrelin injection enhances fat intake over carbohydrate intake
- **Mice** → Increases consumption of palatable saccharin solution and saccharin-flavored foods but not in mice that are GHSR deficient
- **Humans** → Ghrelin administration in humans during fMRI increase neural response to food pictures in brain regions connected to feeding

What does this mean??

Suggests an obligatory role for ghrelin in caloric restriction-associated food-reward behavior
Previous Models: Ghrelin in Stress-Based Eating

- **Humans** → Subjects of the Standardized Trier Social Stress test displayed an increase in plasma ghrelin
- **Mice** → Male mice subjected to Chronic Social Defeat Stress (CSDS) procedure displayed elevated levels of ghrelin
  - After CSDS, most male mice exhibited depression-like behavior
New Model

- Assesses food-reward behavior in mice after exposure to CSDS using CPP task
- Assesses relationship of stress-induced food-reward behavior to rising levels of ghrelin
- Demonstrate:
  - CSDS in mice increases both CPP for HFD
  - Stress-induced food behavior is dependent on ghrelin signaling
  - Ghrelin signaling not only mediates ghrelin's orexigenic, antidepressant-like, and food-reward behavior effect but ALSO stress-induced food-reward behavior
Methods: Animal Models

- 3 Types of Mice
  - GHSR-null or GHSR-deficient
  - GHSR-null/TH
  - Wild-Type
Methods: CSDS and Social Interaction Task

- To induce chronic social defeat stress
  - Based on the Resident-Intruder Paradigm
    - A test mouse was put into the home cage of another aggressive mouse for 5 minutes of physical encounter each day for 10 days, followed by a sensory encounter for the remainder of the day

- Social Interaction Task
  - Conducted on Day 11
  - Involves placing mice in an arena with a small animal cage at one end, with their movement tracked for 2.5 minutes in the absence of another mouse, followed by 2.5 minutes in the presence of caged, unfamiliar target mouse
Methods: CPP

First 12 Days

Odd Days
- Restricted for 20 minutes with an equal calorie amount of regular chow

Even Days
- Spent time in home cage with open access to regular chow

After 12 Days

Free access for 30 minutes to the 2 chambers in the absence of either diet

CPP = Time Spent in Chamber with Regular Chow - Time Spent in Chamber with HFD
Results

**MAIN IDEA:**
Based upon the observations that circulating ghrelin levels rise after various forms of stress, including CSDS; and that ghrelin signaling increases the rewarding properties of energy-dense foods, we hypothesized that *stress-induced elevations in ghrelin mediate reward behaviors aimed at obtaining calorie heavy, pleasureable comfort foods upon social stress*. 
CSDS results in several persisting behavioral deficits reminiscent of depression, including social isolation (Graphs A and B).

CSDS-induced social isolation was intensified by genetic deletion of GHSRs (Graphs A and B).
CSDS resulted in elevations of acyl-ghrelin (confirmed ligand for GHSR), when assessed 1 day after the social interaction test, but not des-acyl-ghrelin (Graphs C and D).
Corticosterone levels measured 1 day after the social interaction test were higher in wild type mice than in those in GHSR-null littermates after CSDS (Graph E)
After CSDS, wild type mice demonstrated an increased preference for the chamber previously paired with HFD (Graph A).

In contrast, neither CSDS-exposed GHSR-null mice nor non-CSDS-exposed control subjects demonstrated CPP for HFD (Graph A).
CSDS-exposed wild type mice gained more weight over the course of the CPP protocol than did control mice or GHSR null mice (Graph B)
The CSDS-exposed wild type mice consumed more HFD during the conditioning sessions than did the other groups (Graph C).
No differences were observed here in consumption of regular chow available during the conditioning session (Graph D)
- Wild type mice, but not GHSR-null mice, responded to injections of ghrelin by increasing their intake of freely available regular chow (Graph B).
- Reexpression of GHSR in TH-containing neurons partially restored ghrelin-induced food intake (Graph B).
- Fasting blood glucose levels were lower in GHSR-null mice than in wild type mice (Graph C).
- However, GHSR-null/TH mice showed similar fasting blood glucose levels to those of GHSR-null mice (Graph C).
- Administered ghrelin induced CPP for HFD (Graph D)
- CSDS induced CPP for HFD (Graph F)
From previous findings, GHSR-null mice showed increased social isolation after CSDS compared to wild type mice.

Reexpressions of GHSR in TH-containing neurons improved social interaction scores to levels similar to that of the wild type mice (Graph E).
Discussion: CSDS

● Why Chronic Social Defeat Stress?
  ○ Representative model of psychosocial stress in humans
  ○ Produces depression state in mice similar to major depressive disorder in humans
  ○ Models changes in eating behavior and body weight gain during stress/depression in humans
Ghrelin’s Role in Stress Eating

- Prolonged psychosocial stress → activate feelings of depression and increases secretion of ghrelin → more ghrelin attaching to GHSR → inhibit feelings of depression, activate the eating of pleasurable foods, increased appetite for food → eat more comfort foods → increased body weight
Additional Information

- Ghrelin levels elevated through an unknown pathway
  - May involve stimulation of $\beta_1$-adrenergic receptors on ghrelin
    - Pathway observed during ghrelin release during 24-hr fast
- Mice lacking GHSRs experienced social avoidance after CSDS
  - Reinforces idea that GHSRs inhibit feelings of depression
- Food-reward behaviors dependent on ghrelin signaling
  - Causes us to seek out comfort foods
How Ghrelin Influences Food Reward Behavior

- Ventral tegmental area (VTA) dopaminergic neurons is a key player in reward behavior
  - GHSRs in VTA dopaminergic neurons
  - Ghrelin act on VTA neurons that influences food reward
    - Ghrelin causes dopamine release in nucleus accumbens
      - ↑ AP frequency in VTA neuron
- GHSRs in substantia nigra dopaminergic neurons
  - Neuronal projection from substantia nigra → dorsal striatum involved in reward behavior
Injections into VTA

- Ghrelin microinjection into VTA
  - ↑ food intake
  - ↑ intake of peanut butter
  - ↑ lever pressing for sucrose reward
- GHSR antagonist microinjection into VTA
  - ↓ food intake
  - ↓ lever pressing for sucrose reward
- VTA lesion
  - ↓ intake of peanut butter
  - ↓ time spent in tubes with peanut butter inside
Ghrelin and Stress Link

- Link between ghrelin, stress, and eating behaviors may have provided survival advantage in past
  - Had to search for food when starving?
  - Store food for winter time?
- Modern society, this link may be more harmful than helpful
  - Lead to obesity in people with major depressive disorder or under chronic stress
- Where to go next?
  - Developing pharmaceutical agents to inhibit this behavior
  - Other hormones involved in stress eating
  - Importance of GHSRs in neurons in substantia nigra and arcuate nucleus
Eating Habits... Do We Have Control?

- Sweet/fatty foods and environmental cues become extremely salient indicators of reward associated with eating
  - Due to evolutionary perspective -> need to keep fat for famine
- In today's day and age.. might lead to overeating of foods not needed for metabolic need = gain weight!
- This increase of salience in food can be mirrored to the rise of addiction to drugs of abuse because it has a neurobiological pathway in common
  - Pathway mediates altered reward salience and motivation -> Mesolimbic DA system!!
Dopamine and Food Addiction

- DA = key player in reward and addiction related behavior
  - Not just about eating, but starving too
  - Genetically altered mice that lack DA signaling = starve
- VTA -> NAc (of ventral striatum) DA pathway extremely important in incentive motivation
- DA neurons activate after food reward, but over time, DA neurons respond more to CUE rather than the actual reward
  - Midterms? Stress?
- Through this mechanism, food cues drive food intake in habitual fashion
  - Do you crave a salad while sitting in Geisel at 3 AM?..... probably not.
- Block DA neurotransmission in NAc = reduced motivated behavior for food
Remember Leptin?

- Domingos et al. (2011)
  - Leptin modulates reward value of sucrose (chief compound of cane/beet sugar)
  - Rats restricted to food intake -> sucrose became higher valued
  - Injected leptin to those restricted rats -> sucrose became less valued
  - Value measured through DA activity
- Cross-over between metabolic signals and neural reward-related signaling!
  - Drives animal to change their motivated behavior in response to fluctuations in energy balance
Eating Addiction? Drug Addiction?

- Rats put under 12h food deprivation followed by 12h intermittent access to normal food and sugar solution
  - Leads to sugar binging
  - Evidence shows endogenous opioid signaling active during sugar binging
  - Injection of opioid receptor antagonist naloxone = somatic effects that are parallel to withdrawal syndrome
- Sugar recruits endogenous opioid pathways and consequences are similar to those that follow taking drugs of abuse
  - However, there has been suggestion that behaviors can also recruit endogenous opioid systems as well
  - Could be a mixture of both?
Eating Addiction? Drug Addiction? (Cont.)

- Food and beverage industry dominated by high-sugar, high-fat products
- Especially as students, we are more attracted to these
  - Quick, on-the-go
  - Cheap
  - Easily accessible
- Continuous consumption of these foods (especially in high-stress environments) creates habits and reinforces faulty reward-prediction behavior
- Creates possibility for becoming addicted to unhealthy foods, in a similar fashion to how one becomes addicted to drugs of abuse
- A huge problem that has existed for ages, something must be done!!
  - Public health, student health advocacy, etc.
Obesity and Interactions with Leptin

Obesity has generated interest in the mechanisms of human metabolism and energy regulation

- Pharmacologic treatments for obesity centers around the key hormonal actions of insulin, leptin, and ghrelin
- Experiments show that leptin and ghrelin have an inverse correlation, but further questions still stand such as...
  - The effect of exogenous leptin or ghrelin in the human model and its role in being a hormonal therapy for obesity
Acknowledgements